8 D
08/951,733
A-4338

Europäisches Patentamt

(19) European Patent Office

Office européen des brevets

(11) Publication number: **0 058 481**
A1

(12) EUROPEAN PATENT APPLICATION

(21) Application number: 82300416.3

(22) Date of filing: 27.01.82

(51) Int. Cl.³: **A 61 K 37/02**
C 08 G 63/08
//C07C103/52, A61K31/765,
A61K9/22

(30) Priority: 16.02.81 GB 8104734

(43) Date of publication of application:
25.08.82 Bulletin 82/34

(84) Designated Contracting States:
AT BE CH DE FR GB IT LI LU NL SE

(71) Applicant: IMPERIAL CHEMICAL INDUSTRIES PLC
Imperial Chemical House Millbank
London SW1P 3JF(GB)

(72) Inventor: Hutchinson, Francis Gowland
29 Woodland Drive
Lymm Cheshire WA13 0BL(GB)

(74) Representative: Atkinson, John David et al,
Imperial Chemical Industries PLC Legal Department :
Patents Thames House North Millbank
London SW1P 4QG(GB)

(54) Continuous release pharmaceutical compositions.

(57) Pharmaceutical compositions, comprising a polylactide and a pharmacologically active, acid stable polypeptide, which when placed in an aqueous physiological environment release the polypeptide at an approximately constant rate in an essentially monophasic manner, with a minimal, or no induction period prior to the release; polylactides suitable for use in said compositions; and a method for the manufacture of such polylactides.

EP 0 058 481 A1

TITLE: CONTINUOUS RELEASE PHARMACEUTICAL COMPOSITIONS

This invention relates to pharmaceutical compositions of pharmacologically-active acid-stable polypeptides, which provide continuous release of the polypeptide over an extended period when the composition is placed in an aqueous, physiological-type environment.

It has long been appreciated that the continuous release of certain drugs over an extended period following a single administration could have significant   practical advantages in clinical practice, and compositions have already been developed to provide extended release of a number of clinically useful drugs, after oral dosing (see, for example, Remington's Pharmaceutical Sciences, published by Mack Publishing Company, Easton, Pennsylvania, U.S.A., 15th Edition, 1975, pages 1618-1631), after parenteral administration (ibidem, pages 1631-1643), and after topical administration (see, for example, United Kingdom Patent Number 1,351,409).   A suitable method of parenteral administration is the sub-dermal injection or implantation of a solid body, for example a pellet or a film, containing the drug, and a variety of such implantable devices has been described.   In particular, it is known that, for many drugs, suitable implantable devices for providing extended drug release may be obtained by encapsulating the drug in a biodegradable polymer, or by dispersing the drug in a matrix of such a polymer, so that the drug is released as the degradation of the polymer matrix proceeds.

- 2 -

Suitable biodegradable polymers for use in sustained release formulations are well known, and include polyesters, which gradually become degraded by hydrolysis when placed in an aqueous, physiological-type environment. Particular polyesters which have been used are those derived from hydroxycarboxylic acids, and much prior art has been directed to polymers derived from α-hydroxycarboxylic acids, especially lactic acid in both its racemic and optically active forms, and glycolic acid, and copolymers thereof - see, for example, United States Patents Numbers 3,773,919 and 3,887,699; Jackanicz et al., Contraception, 1973, 8, 227-234; Anderson et al., ibidem, 1976, 11, 375-384; Wise et al., Life Sciences, 1976, 19, 867-874; Woodland et al., Journal of Medicinal Chemistry, 1973, 16, 897-901; Yolles et al., Bulletin of the Parenteral Drug Association, 1976, 30, 306-312; Wise et al., Journal of Pharmacy and Pharmacology, 1978, 30, 686-689 and 1979, 31, 201-204.

In this specification, the term "polylactide" is used in a generic sense to include polymers of lactic acid alone, copolymers of lactic acid and glycolic acid, mixtures of such polymers, mixtures of such copolymers, and mixtures of such polymers and copolymers, the lactic acid being either in racemic or in optically active form. Also, the term "acid-stable" is to be understood as meaning that the polypeptide is not significantly hydrolysed under the conditions encountered within the claimed formulations during the period of use envisaged, that is, at pH 2 at mammalian body temperature, say up to 40°C., for up to six months.

United Kingdom Patent Specification
Number 1,325,209 (equivalent to United States Patent
Specification Number 3,773,919) and United States
Patent Specification Number 3,887,669 is the only
prior art known to us which makes any reference to
extended or sustained release of polypeptides, the
latter mentioning insulin only, but it contains no
specific example of any such formulation, and the
reference to polypeptides is apparently entirely
speculative, appearing, as it does, only in an
extensive listing of very many different classes of
drugs which can allegedly be incorporated into
formulations of the kind described therein.   In fact,
essentially all of the other drug types referred to in
that specification, apart from polypeptides, are
relatively hydrophobic in character and of relatively
low molecular weight, and the disclosure of that
specification displays no recognition of the difficulties
which we have encountered when seeking to obtain
satisfactory sustained release formulations of
polypeptides, many of which are relatively hydrophilic,
and of relatively high molecular weight.

It is to be appreciated that "sustained" or
"extended" release of a drug may be either continuous
or discontinuous.   We have now discovered, in fact,
that in many cases when the teaching of the prior art,
and in particular the teaching of United Kingdom
Specification No. 1,325,209, is applied to the
manufacture of a formulation of an acid-stable
polypeptide, the release of the polypeptide from the
formulation, although occurring over an extended period
of time, may also be discontinuous.   For example, the
release of a polypeptide from a polylactide polymer as
described in the said Specification is often preceded

by a significant induction period, during which no
polypeptide is released, or is biphasic, and comprises
an initial period during which some polypeptide is
released, a second period during which little or no
5 polypeptide is released, and a third period during which
most of the remainder of the polypeptide is released.
By contrast, it is an object of the present invention
to provide compositions of acid-stable polypeptides
from which, apart possibly from a relatively short
10 initial induction period, the polypeptide is released
continuously, with no periods during which little or
no polypeptide is released.   The words "continuous
release" are used in this specification solely to
describe a release profile which is essentially
15 monophasic, although it may have a point of inflection,
but certainly has no "plateau" phase.

Thus, according to the present invention,
there is provided a pharmaceutical composition
comprising a polylactide, as hereinbefore defined,
20 and an acid-stable polypeptide, which, when placed in
an aqueous physiological-type environment, releases
polypeptide into said aqueous physiological-type
environment in a continuous manner, as hereinbefore
defined, until essentially all of the polypeptide has
25 been released.

This invention is applicable to acid-stable
polypeptides quite generally, without any limitation as
to structure or molecular weight, but is most useful
for polypeptides which are relatively hydrophilic, and
30 the following list, which is not intended to be
exhaustive, is indicative of polypeptides which may be
employed employed in the formulation of this invention:-

oxytocin, vasopressin, adrenocorticotrophic hormone
(ACTH), epidermal growth factor (EGF),
prolactin, luliberin or luteinizing hormone releasing
hormone (LH-RH), insulin, somatostatin, glucagon,
interferon, gastrin, tetragastrin, pentagastrin,
urogastrone, secretin, calcitonin, enkephalins,
endorphins, angiotensins, renin, bradykinin, baci-
tracins, polymyxins, colistins, tyrocidin, grami-
cidines, and synthetic analogues and modifications and
pharmacologically-active fragments thereof.

We have found, however, that polypeptides
which are not stable under acid conditions, are
unsuitable for use in the compositions of this
invention, as they become degraded in the acid
environment produced in the polymer matrix when the
polyactide starts to degrade by hydrolysis, thus
producing carboxylic acid end groups.

By "an aqueous physiological-type environment"
we mean the body, particularly the musculature or
the circulatory system, of a warm-blooded animal,
although in laboratory investigations such an
environment may be mimicked by aqueous liquids,
optionally buffered to a physiological pH, at a
temperature of between 35 and 40°C.

The continuous release composition of the
invention may be placed in the body of an animal which
it is desired to treat with a polypeptide by, for
example, intramuscular or subcutaneous injection, or
by sub-dermal surgical implantation, in conventional
clinical or veterinary manner.

- 6 -

We have discovered that a pharmaceutical composition according to the invention may be designed to provide continuous release of the polypeptide by the appropriate choice or control of various parameters, for example by varying the polylactide composition, particularly the proportion of lactic acid to glycolic acid in copolymers; by controlling the molecular weight of the polylactide, both the weight-average molecular weight, and the molecular weight range or polydispersity, measured by the ratio of the weight-average molecular weight, (M_ω), to the number-average molecular weight (M_n), ie $\frac{M_\omega}{M_n}$; by choice of the proportion of polypeptide to polylactide; or by choice of the geometry of a solid formulation for implantation, or of the particle size in a formulation for injection. The release characteristics of such compositions are also controlled to some extent by the nature of the polypeptide itself. In particular, there is less freedom of choice in defining the parameters mentioned above when designing a composition of the invention for a polypeptide of high molecular weight, (say, greater than 6000), than there is when designing a composition for a polypeptide of lower molecular weight, (say, less than 6000).

We have further discovered that the release of a polypeptide from a composition comprising a polylactide and a polypeptide proceeds by two distinct and independent mechanisms, namely first a diffusion-dependent release of polypeptide out of the polylactide-polypeptide matrix comprising leaching from the surface, and, for low molecular weight polypeptides, some

- 7 -

partition-dependent diffusion of polypeptide per se;
and subsequently, as the polylactide becomes degraded,
diffusion of aqueous polypeptide solution out of the
composition through aqueous channels.

5   In general, the compatibility of polypeptides
in polylactide polymers is limited, except in the case
of low molecular weight (up to, say, 6000 molecular
weight) polypeptides which are capable of having some
specific interaction with the polylactide, for example
10   a low molecular weight polypeptide which is basic, and
which therefore interacts with the terminal carboxylic
acid groups in the polylactide.   Because of this
limited compatibility of polypeptide in polylactide,
a polypeptide/polylactide formulation, when placed in
15   an aqueous environment, releases very little polypeptide
by diffusion through the polymer matrix.   While this
is broadly true for all combinations of polypeptide
and polylactide, matrix diffusion is at a minimum for
high molecular weight polypeptides in high molecular
20   weight polylactides.   Even when some matrix diffusion
resulting in polypeptide release occurs initially on
placing the composition in an aqueous environment,
by release from or very near the surface, this soon ceases
because the diffusion of polypeptide into polylactide
25   is insufficient to result in continuous transport of
polypeptide from inside the composition to its surface.

          When a polypeptide/polylactide composition
is placed in an aqueous environment, water diffuses
into the matrix, and is partitioned between polypeptide
30   and polylactide to form domains of aqueous polypeptide
solution.   This aqueous polypeptide, obtained when
absorbed water is partitioned between polypeptide and

polylactide, is incompatible with, and insoluble in,
polylactides, particularly those of high molecular
weight, so that absorption of water by the composition

5   still further reduces initially any likelihood of
matrix diffusion of polypeptide out of the composition.
If the aqueous domains of polypeptide so formed are
discrete and isolated, then the compositions are
incapable of releasing polypeptide.   However, the
possibility of the aqueous polypeptide domains having

10 some continuity increases with increasing concentration
of polypeptide in the composition, and with increasing
absorption of water, and when the continuity of aqueous
polypeptide domains reaches a sufficient level to
communicate with the exterior surface of the

15 composition, polypeptide starts to be released from
the formulation by diffusion, not through the polylactide
matrix, but through aqueous polypeptide channels.   Even
when some aqueous polypeptide domains near the surface
have extended so as to reach the exterior of the

20 composition, that aqueous polypeptide which exists in
still isolated domains is not released, and is only
released when a secondary hydrophilic path for
diffusion becomes available.   For high molecular weight
polylactides, this secondary hydrophilic diffusion path

25 arises when the polylactide has undergone sufficient
degradation for the rate of absorption of water to
increase significantly.   When this occurs, aqueous pores
or channels are generated in the polylactide matrix, which
allow a continuous and significant release of polypeptide,

30 as aqueous solution, from previously discrete and
isolated domains.

As indicated above, we have discovered that,
when sustained release compositions of the prior art,
and particularly those described in United Kingdom Patent
Specification Number 1,325,209, are used for the release
5   of polypeptides, the initial matrix diffusion phase of
polypeptide release, and the secondary release of
polypeptide consequent upon degradation of the
polylactide, are separated in time with the result
that the release of polypeptide is not continuous, but
10   is biphasic and discontinuous, comprising a small first
release of polypeptide, a dead phase during which
essentially no polypeptide is released, and a subsequent
second release phase, during which substantially all
of the remaining polypeptide is released. We have
15   now discovered that, by appropriate choice of the
parameters of the composition, the matrix diffusion
phase of release, and the subsequent degradation
induced phase of release, can be made to overlap in
time.

20   Thus, according to a further feature of the
invention there is provided a pharmaceutical composition
comprising a polylactide, as hereinbefore defined, and
an acid-stable polypeptide, and exhibiting two successive
phases of release of polypeptide when placed in an
25   aqueous physiological-type environment, the first phase
being release by matrix diffusion and the second phase
being release consequent upon degradation of the poly-
actide, characterised in that the diffusion phase and the
degradation-induced phase overlap in time.

30   The two phases can be made to overlap by
either extending the initial diffusion phase, or making
the degradation-induced phase commence earlier, or both.

The initial matrix release phase is difficult to extend, but is sensitive to the concentration of the polypeptide in the matrix, and to a limited extent, to the nature of the polypeptide, especially its hydrophilicity,

The degradation-induced release phase can be made to start sooner by appropriate choice of polylactide composition (more glycolide-rich polymer molecules, which degrade more quickly than lactide-rich molecules), M_ω (molecules of low molecular weight degrade more quickly to a level at which aqueous channels appear in the matrix), and polypeptide concentration (a higher polypeptide concentration allows more rapid absorption of water, and consequently more rapid generation of continuous aqueous channels which facilitate poly-peptide release).

However, as well as requiring the degradation-induced release phase to start earlier, it is also necessary to control the rate of polypeptide release during this phase, and to ensure that the total duration of this phase is sufficient for its intended clinical or veterinary purpose. One method of extending the duration of the degradation-induced release phase, is to use polylactides which contain lactide-rich molecules, which degrade more slowly than glycolide-rich molecules, or alternatively, polylactides containing molecules of high molecular weight, which take longer to degrade to a level at which aqueous channels are formed, can be used.

- 11 -

It is apparent, therefore, that since glycolide-rich polymer molecules, and/or molecules of low molecular weight, are preferred if the degradation phase is to start quickly, and lactide-rich molecules, and/or molecules of high molecular weight are preferred if the degradation-induced release phase is to last for a sufficient period of time, preferred polylactides are those with a high degree of heterogeneity, in respect of glycolide-rich and lactide-rich molecules, or of high poly-dispersity.

Alternatively, the same characteristics can be obtained by blending two or more different polylactides, which differ in lactide/glycolide content, and/or in M_ω. In addition, the blending of a minor proportion of polylactide of high M_ω with a polylactide of low M_ω, confers desirable physical properties upon compositions according to this invention produced therefrom, making them easier to fabricate and process.

We have further discovered that the profile of polypeptide release from both prior art polylactides and novel polylactides is almost exactly parallelled by the profile of water absorption. That is to say, when polypeptide release is discontinuous, water absorption is also discontinuous in essentially the same manner, and conversely, when polypeptide release is continuous, so is water absorption. Furthermore, variation of the parameters referred to above for controlling the polypeptide release characteristics of the composition have been found to affect water absorption by the composition in an exactly parallel manner.

It is apparent, therefore, that since glycolide-rich polymer molecules, and/or molecules of low molecular weight, are preferred if the degradation phase is to start quickly, and lactide-rich molecules, and/or molecules of high molecular weight are preferred if the degradation-induced release phase is to last for a sufficient period of time, preferred polylactides are those with a high degree of heterogeneity, in respect of glycolide-rich and lactide-rich molecules, or of high poly-dispersity.

Alternatively, the same characteristics can be obtained by blending two or more different polylactides, which differ in lactide/glycolide content, and/or in M_ω. In addition, the blending of a minor proportion of polylactide of high M_ω with a polylactide of low M_ω, confers desirable physical properties upon compositions according to this invention produced therefrom, making them easier to fabricate and process.

We have further discovered that the profile of polypeptide release from both prior art polylactides and novel polylactides is almost exactly parallelled by the profile of water absorption. That is to say, when polypeptide release is discontinuous, water absorption is also discontinuous in essentially the same manner, and conversely, when polypeptide release is continuous, so is water absorption. Furthermore, variation of the parameters referred to above for controlling the polypeptide release characteristics of the composition have been found to affect water absorption by the composition in an exactly parallel manner.

- 12 -

Thus, according to a further feature of
the invention there is provided a pharmaceutical
composition, comprising a polylactide as hereinbefore
defined and an acid-stable polypeptide, which when
5 placed in an aqueous physiological-type environment
absorbs water in a continuous manner, as hereinbefore
defined, until the polylactide has been degraded and
essentially all of the polypeptide has been released
into said aqueous physiological-type environment.

10 The effect of the various parameters,
referred to above, on the polypeptide release and/or
water absorption characteristics of compositions of
the invention is illustrated by the following experiments:-

A. Molecular weight of the polylactide component.
15 A.1. Low molecular weight polypeptide.
A.1.1.Formulations were manufactured comprising
20% w/w of the gastric peptide fragment
tetragastrin hydrochloride, Trp-Met-Asp-
Phe-NH_2.HCl, molecular weight = 633, in a
20 polylactide comprising equimolar proportions
of D,L-lactide and glycolide units, in the
form of a film 0.2mm. thick.   The films
were placed individually in water at 37^OC.,
which was changed daily, and the ultra-
25 violet absorption at 277nm: was measured
to assay the tetragastrin released by the
formulation that day.
With a prior art type of polylactide
of M_ω approximately 240,000 (intrinsic
30 viscosity = 1.36), there was an initial
release of tetragastrin, then a "dead
period" from about day 5 to day 21 during

- 13 -

which little was released, followed by
the main release from day 24 onward.

With a novel polylactide of M_ω approximate-
ly 15,000 (intrinsic viscosity = 0.25), the
release pattern was similar, but the dead
period lasted only from day 4 or 5 to
day 8 or 9.

With a novel polylactide of low M_ω
(inherent viscosity of a 1g./100ml. solution
in chloroform = 0.11), there was no dead
period, and tetragastrin was released
continuously from time zero (T_0).

A.1.2. Similar formulations were manufactured
using 10% by weight of the synthetic
luliberin analogue ICI.118,630, $\llcorner$Glu –His–
Trp-Ser-Tyr-D-Ser(O-tBu)-Leu-Arg-Pro-Azgly-
NH_2, molecular weight = 1269, in place of
tetragastrin.

With a prior art polylactide of M_ω
$\sim$ 240,000 (intrinsic viscosity = 1.36),
the release of the polypeptide was biphasic,
with a dead period of about 15 days.

With a novel polylactide of $M_\omega \sim$ 15,000
(intrinsic viscosity = 0.25), there was
a short induction period, followed by
continuous release.

With a novel polylactide of low M_ω
(inherent viscosity of a 1g./100ml.
solution in chloroform = 0.11), there was
continuous release from T_0.

A.2.   Medium molecular weight polypeptide.
Formulations were manufactured comprising
0.1% by weight of mouse epidermal growth
factor (EGF), molecular weight = 6041, in

- 14 -

polylactides comprising equimolar proportions of D,L-lactic acid and glycolic acid units, and of different M_ω, and placed in pH 7.4 buffer. Release of EGF was monitored by radio-immunoassay.

With a prior art polylactide of $M_\omega \sim 200,000$ (intrinsic viscosity = 1.08), there was no initial release, and no significant release of polypeptide until between 13 and 20 days after T_o, after which release was continuous.

With a novel polylactide of $M_\omega \sim 80,000$ (intrinsic viscosity = 0.58), there was also no initial release, and significant release did not occur until between 6 and 10 days after T_o, after which release was continuous.

With a novel polylactide of low M_ω (inherent viscosity of a 1g./100ml. solution in chloroform = 0.11), there was continuous release from T_o.

A.3. High molecular weight polypeptide.

A formulation was manufactured comprising 20% w/w of bovine prolactin, molecular weight $\sim 22,000$, in a novel polylactide comprising equimolar proportions of D,L-lactic acid and glycolic acid units, and of low M_ω (inherent viscosity of a 1g./100ml. solution in chloroform = 0.11). As expected from previous experiments A.1.1, A.1.2 and A.2, this formulation also released polypeptide continuously from T_o when tested in vivo in rats, and circulating bovine prolactin assayed by radio-immuno assay.

- 15 -

Experiments A.1 to A.3 thus demonstrate that
reducing the molecular weight, M_ω, or viscosity
of the polylactide used in the manufacture of a
composition reduces the biphasicity of poly-
5   peptide release, for polypeptides of low or
medium molecular weight, or the initial delay
in release of polypeptides of medium or high
molecular weight, and achieves a continuous
release of the polypeptide from T_O.

10 B.   Ratio of lactide/glycolide in the polylactide.

Compositions were manufactured in the form
of implants containing 100µg. (3% w/w) of the
luliberin analogue ICI. 118,630 in polylactides
of $M_\omega \sim 300,000$ but of different lactide/
15 glycolide ratios.   All of these formulations
when tested _in_ _vivo_ in adult female rats
exhibiting normal oestrous behaviour, gave
biphasic release of the polypeptide, comprising
release for about 6 days post-treatment, followed
20 by a dead period, during which there was no
significant polypeptide release.   The length of
this dead period decreased with decreasing lactide/
glycolide ratio (L/G), as follows:-

L/G	Dead period (days)
100/0	no release
75/25	51
67/33	34
50/50	15

- 16 -

This experiment thus indicates that a
composition exhibiting a biphasic release
of polypeptide can be improved in the
direction of achieving continuous release

5   by increasing the proportion of glycolide
to lactide in the polylactide used, up to
about 50% glycolide.

C.   Ratio of polypeptide to polylactide.

        Compositions in the form of implants were

10   manufactured using different concentrations of
the synthetic luliberin analogue, ICI.118,630,
in a prior art 50/50 lactide/glycolide
polylactide of $M_\omega \sim$ 200,000, and tested in vivo
in rats as described above. At 5% and 10% w/w

15   incorporation, the release of polypeptide was
biphasic, but at 15% and 20% incorporation, the
biphasicity disappeared.

        This experiment thus demonstrates that
a polylactide of high molecular weight, which

20   gives biphasic release of polypeptide when the
polypeptide is incorporated only at low levels,
can be used to make satisfactory continuous
release formulations if the proportion of
polypeptide is increased sufficiently.

25   D.   Molecular weight distribution.

        A solution of a polymer blend of wide
molecular weight distribution (polydispersity) was
obtained by mixing solutions of a 50/50 D,L-
lactide/glycolide polylactide of low M_ω,

30   (reduced specific viscosity of a 1g./100ml.
solution in chloroform = 0.115), (3 parts by
weight) and a 50/50 D,L-lactide/glycolide
polylactide of M_ω = 200,000, (intrinsic viscosity
= 1.08), (1 part by weight). Tetragastrin

(1 part by weight) was added, and the
mixture was cast to give a polylactide/
tetragastrin composition containing
20% by weight of tetragastrin, and this was then
moulded to give a slab 0.02cm. thick.   The slab
was placed in water at 37°C.   and the release of
tetragastrin was found to be continuous from T_O,
and to continue for at least 44 days.

Polylactides of wide molecular weight
distribution may be obtained either by mixing
preformed polymers of different molecular weights,
or by appropriate control of the polymerisation
process in generally known manner, and such
polylactides confer important advantages, for
example the lower molecular weight polylactide
species allow an essentially immediate release of
some polypeptide, while the higher molecular.
weight polylactide species both extend the
release period and slow down the overall rate of
release of the polypeptide.   In addition, the
blending of low and high M_ω fractions modifies
the water absorption characteristics of the
polylactide in a parallel manner.

E.   Thickness of the implant.

E.1.   A solution of 10% by weight of tetragastrin
        in a polylactide comprising equimolar
        proportions of D,L-lactic acid and glycolic
        acid units, and of $M_\omega \sim$ 15000 was cast
        into films 0.02, 0.06 and 0.12 cm. thick.
        All three films showed continuous release of
        tetragastrin from T_O, and at 28 days, the
        three films had released respectively 85,
        75 and 66% of their tetragastrin content.

E.2.   The uptake of tritiated water from pH 7.4 buffer by slabs of polylactide comprising equimolar proportions of D,L-lactic acid and glycolic acid units of $M_\omega \sim 200,000$, and intrinsic viscosity = 1.08 and of thickness 0.02, 0.06, 0.12 and 0.20 was measured by removing such slabs from the buffer solution successively after varying times of immersion, and measuring the tritium content by scintillation counting.   After 5 weeks, the different thickness slabs had absorbed respectively 44, 20, 15 and 11% by weight of water.

These experiments show how the thickness of the implant can be used to control the absorption of water by, and thus the rate of release of polypeptide from, a composition of the invention, thicker implants releasing the polypeptide more slowly than thinner ones.

As indicated above the composition of the invention may be formulated as a solid composition for sub-dermal injection or implantation or as a liquid formulation for intramuscular or subcutaneous injection.

Suitable solid compositions for sub-dermal injection or implantation are, for example, rods, spheres, films or pellets, and cylindrical rods which can be injected through a hypodermic needle or trochar are preferred.   Such compositions are manufactured by conventional techniques which are well known in the pharmaceutical art.

- 19 -

Preferred solid compositions of the invention, for polypeptides within different molecular weight ranges, are as shown in Table 1, and preferred compositions for specific polypeptides of particular interest are as shown in Table 2. Each entry in Tables 1 and 2 thus defines a further feature of this invention.

TABLE 1

No.	Polypeptide M.W.	Polylactide Inherent Viscosity	Glycolide Lactide	% Poly-peptide	Preferred % Poly-peptide
1.	<2000	>0.5	0.5-3	5-50	
2.	<2000	0.2-0.5	0.2-3	5-50	10-30
3.	<2000	<0.2	0-3	0.1-50	
4.	1500-10,000	0.4-0.8	0.5-3	10-50	20-50
5.	1500-10,000	0.15-0.4	0.2-3	5-30	10-30
6.	1500-10,000	<0.15	0-3	0.1-20	1-20
7.	8000-30,000	0.15-0.4	0-3	0.1-50	
8.	8000-30,000	0.1-0.15	0.7-3	10-50	
9.	8000-30,000	<0.1	0-3	0.1-50	

- 20 -

TABLE 2

No.	Polypeptide	Polylactide Inherent Viscosity	Glycolide Lactide	% Poly-peptide
10.	Tetragastrin	>0.5	1-3	5-50
11.	Tetragastrin	0.2-0.5	0.5-3	5-50
12.	Tetragastrin	<0.2	0-3	0.1-50
13.	ICI.118630	>0.5	0.8-3	5-50
14.	ICI.118630	0.2-0.5	0.2-3	5-50
15.	ICI.118630	<0.2	0-3	0.1-50
16.	EGF	0.4-0.8	0.5-3	10-50
17.	EGF	0.15-0.4	0-3	0.1-50
18.	Prolactin	<0.15	0-3	0.1-50

Also as indicated above, the composition of
the invention may also be formulated as a suspension for
injection.   Such suspensions may be manufactured by
5   general techniques well known in the pharmaceutical
art, for example by milling the polylactide/polypeptide
mixture in an ultracentrifuge mill fitted with a
suitable mesh screen, for example a 120 mesh, and
suspending the milled, screened particles in a
10   solvent for injection, for example propylene glycol,
water optionally with a conventional viscosity increasing
or suspending agent, oils or other known, suitable
liquid vehicles for injection.

The continuous release of ICI.118630 from a
suspension composition of the invention was demonstrated
by comparing the dioestrous behaviour of normal mature
female rats dosed subcutaneously with either a

5   propylene glycol suspension of particles milled to
120-mesh size and containing 3% by weight of ICI.118630
in a polylactide comprising equimolar proportions of
D,L-lactic acid and glycolic acid units, and a $M_{\omega} \sim$
240,000, (intrinsic viscosity = 1.36), or with a saline

10 solution of 100, 200 or 300µg. of ICI.118630 per animal.
With the saline solutions, there was an immediate short
period of dioestrus, but after 3 days post-dosing normal
cyclicity was re-established.   With the suspension
composition of the invention, by contrast, the animals

15 were substantially in dioestrus for about 40 days.
Similar results were obtained with an injection
formulation based on a mixture of 1% of ICI.118630 in
a similar polylactide having an intrinsic viscosity of
> 0.5.

20   Thus, according to a further feature of the
invention there is provided a suspension formulation
comprising from 1 to 50% by weight of a solid formulation,
which itself comprises from 0.1 to 50% by weight of an
acid-stable polypeptide as herein defined and from 50 to

25 99.9% by weight of a polylactide wherein the ratio of
glycolide to lactide units is 0 to 3 and which is either
soluble in benzene and has an inherent viscosity (1g./
100ml. solution in benzene) of less than 0.5 or is
insoluble in benzene and has an inherent viscosity (1g./

30 100ml. solution in chloroform or dioxan) of less than 4,
which solid formulation has been reduced to fine particle
size, together with from 50 to 99% by weight of a liquid
carrier suitable for injection into mammals.

It is to be noted that, because of the
reduced particle size of the polypeptide/polylactide in
a suspension for injection, certain solid formulations,
which are not suitable for implantation, are rendered
useful when reduced to fine particle size and formulated
as a suspension for injection. For example, the two
particular suspension formulations referred to above
both contain less ICI.118,630 than has been found
acceptable for implantable formulations, as shown in
Tables 1 and 2 above.

From the foregoing, it is clear that it is
desirable to manufacture polylactides of a range of
M_ω , particularly of low to medium M_ω in the range
up to 60,000, and of high polydispersity $\left(\dfrac{M_\omega}{M_n}\right)$, these

being particularly valuable in the compositions of this
invention.   The prior art relating to polylactides in
general, and to copolymers containing lactic acid and
glycolic acid units in particular, is silent as to the
manufacture of such copolymers of low molecular weight,
and as to methods of achieving high polydispersity in
such copolymers.   Indeed, it is inherent in the prior art
disclosure of polylactides that they are generally of M_ω
greater than about 30,000-60,000 (an inherent viscosity of
greater than 0.5) and of low polydispersity, due to
their manufacture under anhydrous conditions and
without any added chain-stoppers.   We have realised
that, because of the different reactivities under
polymerisation conditions of the cyclic dimers of
lactic acid and glycolic acid, copolymers of high
heterogeneity in respect of polymer species may be
obtained by ring opening polymerisation of a mixture
of the two cyclic dimers in the presence of chain-

stopping agents, to give polylactides having an
inherent viscosity of less than O.5.   The cyclic
dimer of glycolic acid is the more reactive under
polymerisation conditions, and thus the first copolymer
molecules formed in the polymerisation are glycolic
acid-rich.   Consequentially, the later copolymer
molecules formed are necessarily lactic acid-rich,
thus producing a copolymer of lactic acid and glycolic
acid of the desired high heterogeneity. .

In addition, we control the polymerisation
to produce copolymers in the desired low M_w range by
carrying out the ring opening copolymerisation of
the mixed cyclic dimers in the presence of water,
of lactic acid containing water, or of some other
known chain growth regulator, in accordance with
the general knowledge in the polymer art.

Suitable polymerisation catalysts are zinc
oxide, zinc carbonate, basic zinc carbonate, diethyl-
zinc, organotin compounds, for example stannous
octanoate, tributylaluminium, titanium, magnesium
or barium compounds, or litharge, and of these
stannous octanoate is preferred.

The copolymerisation of the mixed cyclic
dimers is otherwise carried out in conventional
manner, known in the polymer art, as regards time
and temperature. .

Low molecular weight polylactides may also be
obtained by copolymerisation of the hydroxy-acids
themselves rather than the cyclic dimers.   Although
less heterogenerous polymers are obtained by this
process, suitable matrices for continuous release of

- 24 -

polypeptides may be obtained by mixing such polylactides of different compositions made by this method, or by mixing a polylactide made by this method with one or more polylactides made by ring opening polymerisation of the cyclic dimers.

Certain of the lactic acid/glycolic acid copolymers described herein are novel and form a further feature of this invention. Thus according to a further feature of the invention there is provided a heterogeneous polylactide comprising from 25 to 100% molar lactic acid units and from 0 to 75% molar of glycolic acid units, and which is either soluble in benzene and has an inherent viscosity (1g./100ml. solution in benzene) of less than 0.5, or is insoluble in benzene and has an inherent viscosity (1g./100ml. solution in chloroform or dioxan) of less than 4. By a "heterogeneous polylactide" we mean polylactides with a high degree of heterogeneity, in respect of glycolide-rich and lactide-rich molecules, or of high polydispersity, or blends of two or more different polylactides which differ in lactide/glycolide content and/or M_ω, as hereinbefore described.

Whether or not a particular copolymer is heterogeneous or not, in this sense, may be readily deterimined from inspection of the 25MHz ^{13}C nuclear magnetic resonance spectrum of the copolymer in, for example, deuterated dimethyl sulphoxide. In a homogeneous copolymer, such as is obtained in the prior art copolymerisation of lactic acid and glycolic acid monomers, the resonance of the glycolic acid unit carbonyl carbon, at $\delta = 166.0 - 166.2$ approximately, appears as two doublets, as a consquence of the four different, approximately equally probable molecular environments in which this carbon atom can exist,

namely GĠG, LĠG, GĠL and LĠL (G = a glycolic acid unit,
L = a lactic acid unit, and the asterisk indicates the
glycolic acid unit under consideration). In a hetero-
geneous copolymer, on the other hand, such as is used in
the present invention, the sequence LĠL is unlikely to
occur, so that one of the doublet singnals in the spectrum
of the homogeneous copolymer appears as a singlet. In
fact, we find that in the spectrum of heterogeneous
copolymers, this glycolic acid unit carbonyl carbon
signal often appears as two singlets. Thus, a
"heterogeneous copolymer" as herein defined is a copolymer
for which the glycolic acid carbonyl carbon signal in the
^{13}C n.m.r. appears as other than a pair of doublets.

The heterogeneity or homogeneity of lactic
acid/glycolic acid copolymers can also be demonstrated
by an examination of their degradation. Thus, when a
copolymer is placed in pH 7.4 buffer at 37°C., removed
periodically, dried and sampled, and the ratio of lactic
acid to glycolic acid units in the samples is determined
by n.m.r., for a heterogeneous copolymer, the ratio L/G
increases with time, as the glycolic acid sequences
hydrolyse preferentially. For a homogeneous copolymer,
on the other hand, the ratio of L/G remains essentially
constant as degradation progresses.

The lactic acid content of the copolymer is
preferably in the racemic (D,L) form, or in the
optically active L form.

According to a further feature of the invention
there is provided a process for the manufacture of a
novel copolymer of lactic acid and glycolic acid as
defined immediately above, which comprises the ring
opening copolymerisation of a mixture of the cyclic

dimers of lactic acid and glycolic acid, optionally in the presence of a chain growth regulating agent.

A suitable chain growth regulating agent is, for example, water, lactic acid, glycolic acid or other hydroxy acids, alcohols or carboxylic acids generally.

The invention is illustrated but not limited by the following Preparations and Examples:-

Preparation 1

Zinc oxide (16g.) was added to D,L-lactic acid (800g.) in a 2 1. 3-necked round bottom flask-equipped with a stirrer, a thermometer, and a distillation head connected to a water condenser. The mixture was stirred and heated to about 135°C., at which temperature water started to distil over. Heating was continued for 8 hrs., during which time the temperature rose to about 190°C. When distillation of water ceased, the pressure was reduced, and distillation was continued until solid began to collect in the condenser. At this stage the water condenser was replaced by an air condenser, and the residue was cooled and then distilled under high vacuum (2-8mm. of mercury), the fraction (about 300g.) distilling between 130 and 160°C. being collected, this being D,L-lactide (3,6-dimethyl-1,4-dioxan-2,5-dione), the cyclic dimer of D,L-lactic acid.

The crude D,L-lactide was crystallised from ethyl acetate (approximately 600ml.) three times, and the recrystallised product was finally dried at 45°C. under reduced pressure (2mm. of mercury) for 24-48 hours, after which it had m.p. 124-125°C.

Preparation 2

Glycolide (1,4-dioxan-2,5-dione), the cyclic dimer of glycolic acid, was prepared by the method described in Preparative Methods in Polymer Chemistry by W.R. Sorenson and T.W. Campbell, second edition, published by Interscience (1968), page 363. The crude glycolide was purified by three successive crystallisations from dry ethyl acetate, then dried at 45°C. under reduced pressure (2-8mm. of mercury) for 24-48 hrs., m.p. 82-84°C.

Examples 1 to 13

Polymers of D,L-lactide and glycolide were prepared as follows:-

Pure dry D,L-lactide (Preparation 1), pure dry glycolide (Preparation 2) totalling 42g., commercial D,L-lactic acid containing about 12% by weight of water, and 1ml. of an 8% by weight solution of stannous octanoate in hexane, were placed in a pre-dried glass tube. The hexane was evaporated under reduced pressure, and the tube was heated at 160°C. for 6 hours with constant agitation if possible. The tube was cooled in powdered solid carbon dioxide, and the polylactide was removed, broken into small pieces and dissolved in chloroform (400ml.). The chloroform solution was filtered, and the filtrate was poured into methanol (2 1.) to precipitate the polylactide, which was filtered off and dried under vacuum at 40°C. for 24 hours, then at 80°C. for 24 hours. All the polylactides so produced were soluble in chloroform and dioxan, and polylactides 1 to 9 in the following Table were soluble in benzene, but polylactides 10 to 13 were insoluble in benzene.

- 28 -

The following particular polylactides were prepared by this method:-

Ex.	D,L-Lactide (L) (g.)	Glycolide (G) (g.)	L/G Molar Proportion	D,L-Lactic acid	Intrinsic Viscosity	M_ω (approx)
1	42.0	0	100/0	0	1.385	440,000
2	33.5	9.0	75/25	0	1.084	400,000
3	32.4	8.7	75/25	920µl.	0.108*	low
4	30.0	12.1	67/33	0	0.97	370,000
5	30.0	12.1	67/33	0	0.94	214,000
6	30.0	12.1	67/33	30µl.	0.67	107,000
7	30.0	12.1	67/33	60µl.	0.51	63,000
8	30.0	12.1	67/33	120µl.	0.37	33,000
9	30.0	12.1	67/33	920µl.	0.121*	low
10	23.0	18.5	50/50	0	1.045	300,000
11	23.0	18.5	50/50	400µl.	0.25	15,200
12	23.0	18.5	50/50	920µl.	0.126*	low
13	23.0	18.5	50/50	1380µl.	0.108*	low

M_ω are relative to polystyrene standard

*   are reduced specific viscosities of a 1g./100ml. solution in chloroform.

Alternatively, the lactide, glycolide and lactic acid if present, may be heated at 160°C. and 0.08g. of stannous octanoate then added to initiate the polymerisation.

Example 14

A polylactide comprising equimolar proportions of glycolic acid and D,L-lactic acid units, and having an intrinsic viscosity of 1.36 (50mg.) was dissolved in dioxan (1ml.), and 50μl. of a solution of ICI.118630, $\overline{\text{Glu}}$-His-Trp-Ser-Tyr-D-Ser(O-tBu)-Leu-Arg-Pro-Azgly-NH$_2$ (233mg. per ml. of the acetate salt, equivalent to 200mg. per ml. of base) in distilled water was added. The resultant hazy solution was cast as a film, the solvents were evaporated in a stream of nitrogen in the dark, and the film was dried at 40°C. under reduced pressure (0.02mm. of mercury) for 48 hours. The mixture, containing ~ 17% by weight of ICI.118630 in the polylactide was homogenized by three successive compression mouldings at 110°C. for 10 seconds, and was finally compression moulded into implants 0.038cm. thick, each weighing 1.5mg. and containing 309±7μg. (~ 17% by weight) of ICI.118630,

The continuous release of ICI.118630 from such implants was demonstrated by placing them in female rats exhibiting normal oestrous behaviour. After implantation, the rats went into a period of dioestrus, detected by the lack of cornified vaginal smears, lasting from 31 to 40 days, thus indicating that ICI.118630 was being continuously released during that period.

The process described above was repeated, using 50μl. of ICI.118630 acetate solution (150mg. of pure peptide per ml. of water) and implants were made

- 3c -

similarly, weighing 2mg., containing $306^{\pm}6$ µg. of
ICI.118630 (13% by weight), and 0.038cm. in thickness.
In the rat oestrus test described above, these implants
released ICI.118630 continuously over a period of
5   30 to 38 days, as evidence by the period of dioestrus
in the rats.

For use in human therapy, implants containing
1 to 100mg. of ICI 118630 (5-50% by weight), weighing
2mg.-1g., and in the form of cylindrical rods suitable
10   for implantation by trochar, were manufactured by the
process described above.

Example 15

The process described in Example 14 was
repeated, but using polylactides comprising equimolar
15   proportions of D,L-lactic acid and glycolic acid units,
and having intrinsic viscosities of 0.33 and 0.25,
instead of 1.36, to prepare implants containing 10%
by weight of ICI.118630, weighing about 3mg., and 0.08cm.
thick.

20   These implants were placed in female rats
(5 per group) which, prior to implantation, exhibited
regular oestrous behaviour.   The implants prepared from
polylactide of intrinsic viscosity 0.33 exhibited an
induction period of 5 days followed by a dioestrus
25   period of about 26 days; and the implants prepared
from polylactide of intrinsic viscosity 0.25 exhibited
an induction period of 3-4 days, followed by a dioestrus
period of about 25 days.

Similar implants, but containing 20% by
30   weight of ICI.118630 were prepared in the same way, and
these exhibited a similar dioestrus period, but no
induction period, in the rat test described above.

For use in human therapy, implants containing
1 to 100mg. of ICI.118630 (5-50% by weight), weighing
2mg.-1g., and in the form of cylindrical rods suitable
for implantation by trochar were manufactured similarly.

Example 16

The process described in Example 14 was
repeated, using a polylactide comprising equimolar
proportions of D,L-lactic acid and glycolic acid units,
and having intrinsic viscosity of 1.36, to prepare
mixtures of ICI.118630 and polylactide containing 3%
and 1% by weight of ICI.118630. The mixture was
micronised at room temperature in an ultracentrifuge
mill fitted with a 120-mesh screen, and the micronised
particles were suspended in propylene glycol for
injection at a concentration of 100mg. per ml.

Female rats showing regular oestrous
behaviour were injected sub-cutaneously with 0.1ml.of
the 3% by weight propylene glycol suspension described
above, or with 0.3ml. of the 1% suspension. Both
groups were monitored, by examining vaginal smears
daily for cornification, and exhibited occasional
cornified smears up to 20 to 24 days after dosing,
followed by a clear dioestrus period up to days 38 to
42 after dosing, thus demonstrating continuous release
of ICI.118630 over that period.

Example 17

Tetragastrin hydrochloride, (Trp-Met-Asp-Phe-
NH_2.HCl), (200mg.) was dissolved in a mixture of dioxan
(9ml.) and water (1ml.), and to the solution was added
a polylactide as described in Example 11 (1.8g.). The

mixture was cast as a film, and the solvents were evaporated in a stream of nitrogen. The resulting film was dried under reduced pressure (0.02mm. of mercury) at 40°C. for 48 hours, then homogenised by three successive compression mouldings at 80°C. for 10 seconds, and moulded to give films of thickness 0.02, 0.06 or 0.12cm., each weighing about 80mg.

The release of tetragastrin was measured by placing a film in distilled water, removing a sample of the distilled water daily, replacing all the remaining distilled water by fresh, and measuring the ultra-violet absorption of the daily samples at 277nm. The following results were obtained, which demonstrated continuous release of tetragastrin from films of all three thicknesses:-

Time (days)	Cumulative % Tetragastrin Released		
	0.02cm. Film	0.06cm. Film	0.12cm. Film
1	9.6	5.6	4.0
4	14.9	10.5	9.0
7	20.3	13.9	11.7
9	25.3	17.7	14.8
11	33.1	22.6	19.4
14	48.6	33.2	28.1
17	61.9	45.9	40.5
21	74.7	59.8	53.2
24	81.8	68.1	60.2
28	85.2	74.9	66.4
31	86.9	77.7	70.9
36	88.5	82.5	77.5
39	89.2	85.1	82.6

Example 18

Tetragastrin hydrochloride (40mg.) was dissolved in aqueous dioxan (1:9 by volume), and added to a solution of:

5   (a)   a polylactide comprising equimolar proportions of D,L-lactic acid and glycolic acid units, and having a reduced specific viscosity of 0.115 (as a 1g./100ml. solution in chloroform), (120mg.), and

10 (b)   a polylactide comprising equimolar proportions of D,L-lactic acid and glycolic acid residues, and having an intrinsic viscosity of 1.08 (40mg.), in dioxan (2ml.). The mixed solutions were cast as a film, as
15   described in Example 14, and moulded as implants weighing about 50mg., and 0.02cm. thick.

The release of tetragastrin from these implants was measured by the procedure described in
20 Example 17, and the following results showing continuous release were obtained:-

Time (days)	Cumulative % Tetragastrin Released
1	0.6
2	1.0
3	1.6
4	2.7
7	8.6
10	17.2
14	29.4
18	43.1
23	56.3
28	64.9
32	71.2
36	79.0
39	83.9
44	90.5

Example 19

A polylactide comprising equimolar proportions of D,L-lactic acid and glycolic acid units, and having an inherent viscosity of 0.11 as a 1g./100ml. solution in chloroform, (50mg.), was dissolved in dioxan (1ml.) and a solution of mouse epidermal growth factor (EGF, 0.05mg.) in water (0.05ml.) was added. The mixture was cast as a film on polytetrafluoroethylene cloth, and the solvent was removed in a stream of nitrogen in the dark. The film was dried at 60°C. under reduced pressure (0.8mm. of mercury) for 48 hours. The film was then compression moulded at 120°C. for 10 seconds to give implants 0.02cm. thick, weighing 10mg.

- 35 -

The implants were each placed in a black vial at 37°C. with 1ml. of McIlvain's buffer (pH 7.4), (Documenta Geigy, Scientific Tables, edited by K.Diem and C. Leutner, published by J.R. Geigy SA, Basle, Switzerland, 7th Edition, 1970, page 280), containing 0.02% by weight of sodium azide. The buffer was removed daily, and replaced with fresh, and the EGF released into the buffer by the implant was measured by radio-immuno assay, which demonstrated that release started immediately, and continued for at least 2 weeks releasing 100-200µg. per day.

Example 20

A polylactide comprising equimolar proportions of D,L-lactide and glycolide and having a reduced specific viscosity (1g./100ml. solution in chloroform) of 0.11, (400mg.), was dissolved in dioxan (2ml.), and a solution/suspension of bovine prolactin (100mg.) in distilled water (0.5ml.) was added, with vigorous agitation. The mixture was poured onto a polytetra-fluoroethylene cloth, and dried, first in a stream of nitrogen and then under reduced pressure (0.01mm. of mercury) at 40°C. for 24 hours. The heterogeneous mixture thus obtained was homogenised by four successive compression mouldings at 60°C., and then moulded into a slab 0.2cm. thick, from which implants weighing 60mg. were excised.

The implants were place subcutaneously into adult female rats, which were then periodically bled from the tail, and the prolactin in the blood samples thus obtained was measured by radio-immune assay. A placebo group, receiving no prolactin in

the implant, was similarly assayed, and the levels
of circulating prolactin compared.   The following
results were obtained:-

| Time | Plasma level of bovine prolactin (µg./ml.) | |
	Placebo group	Treatment group
1	0.38	24.7
2	0.45	105.9
6	0.54	7.7
9	0.72	17.8
13	0.52	65.4
16	0.56	89.7
20	0.75	288
23	0.81	142
26	0.84	562
42	1.25	1250

5   Example 21

The process described in Examples 1-13 was
repeated, except that the polylactide was dissolved
in dioxan instead of in chloroform, and similar
polylactides were obtained.

10   Examples 22-29

The process described in Examples 1-13 was
repeated, except that the polylactide was dissolved in
glacial acetic acid, and the glacial acetic acid solution
thus obtained was added dropwise to methanol in order

- 37 -

to precipitate the polylactide, which was filetered off and dried under vacuum at 40°C. for 24 hours, then at 80°C. for 24 hours.

The following particular polylactides were
5   prepared by this method:-

Ex	D,L-lactide (L) (g)	Glycolide (G) (g)	L/G Molar proportion	D,L-Lactic acid	Intrinsic Viscosity	Mw. (approx.)	stannous octanoate
22	33.3	26.7	50/50	360µl	0.234	13,500	93µl
23	11.1	8.9	50/50	120µl	0.243	14,450	31µl
24	11.1	8.9	50/50	120µl	0.265	16,800	31µl
25	111.0	89.0	50/50	1.2ml	0.257	15,850	0.31ml
26	111.0	89.0	50/50	1.2ml	0.239	14,200	0.31ml
27	88.8	71.2	50/50	0.96ml	0.262	16,600	0.25ml
28	22.2	17.8	50/50	1.38ml	0.09*		62µl
29	20.0	-	100/0	0.12ml	0.260	16,200	31µl

* Inherent viscosity as 1gm./100ml. solution in
  chloroform.

Example 30

A polylactide comprising equimolar proportions
10   of glycolic acid and D,L-lactic acid units and having an intrinsic viscosity of 0.25 was dissolved in glacial acetic acid, and the solution was freeze-dried. The freeze dried powder (540.7mg.) and ICI 118630 (142.1mg.) of the acetate salt (equivalent to ≥ 124mg. of base)
15   were dissolved in 6.8ml. of acetic anhydride-free glacial acetic acid and freeze dried for 24hrs.

- 38 -

(The glacial acetic acid was refluxed for 2 hrs. with 1%
water to remove the acetic anhydride): The freeze
dried product was extruded under pressure at 70°C.
to a 1mm. diameter rod, from which implants of the
5   required weight were cut. The implants were dissolved
in an appropriate solvent, for example acetonitrile,
and assayed for drug content and purity. Implants
were shown to contain 16.1% w/w pure 118630 base.

Release of 118630 was evaluated by immersing
10   implants weighing approximately 10mg. in McIlvains pH 7.4 buffer at
37°C. ICI 118630 was released continously for at least
5 weeks.

In a further experiment, implants weighing
approximately 390µg., 860µg., 1500µg., 3000µg and
15   ∼ 6000µg were implanted subcutaneously in groups of
adult, regularly cycling female rats. In the 28 days
following implantation, animals were essentially free
of oestrus intervals showing that active drug was
released continuously over this period.

20   Example 31

A solution of ICI 118630 acetate salt was
prepared by dissolving 170.8mg. of ICI 118630 acetate
in 5ml. of acetic anhydride-free glacial acetic acid.
(The glacial acetic acid was fluxed for 2 hrs. with 1%
25   water to remove acetic anhydride). This solution was
shown by high pressure liquid chromatography (HPLC),
to contain 25.21mg. of ICI 118630 base per ml. 442.5mg.
of polylactide (prepared as in Example 25 was dissolved
in 4.5ml. of the acetic acid solution, and the
30   resulting solution was freeze dried for 25 hrs. The
freeze dried product was extruded under pressure at

74°C. to a 1mm. diameter rod, from which implants of the required weight were cut. The implants were dissolved in an appropriate solvent, such as acetonitrile, and the resulting solution was analysed by HPLC. The implants were shown to contain 20% w/w pure 118630 base.

Example 32

A polylactide comprising equimolar proportions of D,L-lactic acid and glycolic acid units and having a reduced specific viscosity of 0.126 (1g./100ml. solution in chloroform), (240mg.), was dissolved in glacial acetic acid (5ml.) and a solution of tetragastrin hydrochloride (60mg.) dissolved in glacial acetic acid (5ml.) was added with vigorous agitation. The solution was freeze dried for 24 hrs. and the resultant solid was compression moulded at 50°C. for 20 seconds to give implants 0.2cm thick and weighing 35-40mgs.

The above procedure was repeated with the following polymers:-

(a)   a polylactide comprising 67 mole % of D,L-lactic acid and 33 mole % glycolic acid units and having a reduced specific viscosity of 0.121 (1g./100ml. solution in chloroform).

(b)   a polylactide comprising 75mole % of D,L-lactic acid and 25mole % glycolic acid units and having a reduced specific viscosity of 0.108 (1g./100ml. solution in chloroform).

(c)   a polylactide comprising 100% of D,L-lactic acid and having a reduced specific viscosity of 0.100 (1g./100ml. solution of chloroform).

- 40 -

The release of tetragastrin from these implants was measured by the procedure described in Example 17, and the following results were obtained:-

Time (days)	Cumulative % Tetragastrin released			
	50% D,L-lactide 50% glycolide	67% D,L-lactide 33% glycolide	75% D,L-lactide 25% glycolide	100% D,L-lactide
1	6.0	1.0	1.7	1.9
3	12.9	2.1	3.2	3.0
7	23.3	7.1	8.0	5.1
9	27.2	12.2	11.6	6.5
11	30.3	20.2	15.8	8.2
15	36.7	40.0	27.0	14.0
17	39.3	45.0	29.7	17.9
21	44.5	51.8	35.1	25.5
24	49.6	55.6	37.9	30.4
28	58.8	59.6	41.1	36.1
31	68.8	62.3	42.8	40.1
35	81.5	67.3	45.0	45.6
39	91.0	74.3	47.6	50.7
42	95.9	81.9	50.6	55.3
46	96.5	89.1	55.5	60.4
49	97.5	93.2	60.0	65.2
53		95.4	64.8	70.0
56		96.3	68.6	73.6
59		97.0	73.1	77.8
63		97.2	77.1	81.5
70			82.7	86.1
74			85.0	87.5
84			90.4	89.5

These results show that continuous release of
tetragastrin is obtained using low molecular weight
polylactides and that duration of release is determined
by the composition of the hydrolytically unstable
5   polyester.

Example 33

        A polylactide comprising equimolar proportions
of D,L-lactic acid and glycolic acid units and having
an inherent viscosity of 0.126 (as a lg./100ml. solution
10   in chloroform) (9.5mg.), was dissolved in distilled
dioxan (0.25ml.) and a solution of mouse epidermal
growth factor (EGF 0.5mg.) in distilled water (0.1ml.)
was added.   The mixture was cast as a film on poly-
tetrafluoroethylene cloth, and the solvent was removed
15   in a stream of nitrogen, in the dark.   The film was
dried at 40°C. under reduced pressure (0.01mm. of
mercury) for 48 hrs.   The film was then compression
moulded at 70°C. for 10 seconds to an implant 0.02cm.
thick, weighing about 9mgs.   A placebo implant was
20   also prepared.

        The samples were implanted subcutaneously
into carotid cannulated guinea pigs, blood samples
taken periodically and the plasma EGF levels were
measured by radio-immuno assay.

25   Raised plasma EGF levels were observed from
Day 3 and continued for at least 1 week.

        Similar implants were prepared as described
above, but using a polylactide comprising equimolar
proportions of D,L-lactic acid and glycolic acid units
30   and having an intrinsic viscosity of 1.06.   Compression

- 42 -

moulding of this implant was done at 120°C.

Implantation and plasma assays were done as described above, but raised plasma EGF levels were not observed until day 17 after implantation.

5   Example 34

A polylactide comprising equimolar proportions of D,L-lactic acid and glycolic acid units and having an inherent viscosity of 0.093 as a 1g./100ml. solution in chloroform (40mg.) was dissolved in anhydrous-free
10   glacial acetic acid (1ml.) and a solution of mouse epidermal growth factor (EGF, 8.15mg.) in a mixture of water (0.5ml.) and anhydride-free glacial acetic acid (3ml.) was added.   The solution was freeze-dried for 24 hrs. · The resulting powder was then compression
15   moulded at 50°C. to give an implant 2mm.x 2mm.x 10mm. weighing 36.1mg.

The sample was implanted subcutaneously into a cannulated cat, blood samples were taken periodically and the plasma EGF levels were measured
20   by radio-immuno assay.

Raised plasma EGF levels were observed from day 3 and continued for at least 40 days.

Example 35

Implants containing bovine prolactin were
25   prepared as described in Example 20, but using:-

(a)   a polylactide (400mg.) comprising equimolar proportions of D,L-lactic acid and glycolic acid units

- 43.-

and having a reduced specific viscosity (1g./100ml.
solution in chloroform) of 0.11, dissolved in 4ml
dioxan; and

(b)   a polylactide (400mg.) comprising equimolar
5. proportions of D,L-lactic acid and glycolic acid units
and having an intrinsic viscosity of 1.06 dissolved
in 4ml. dioxan.   This sample was moulded at 110°C.

Formulations (a) and (b) were each tested
in vivo as described in Example 20.   Formulation (a)
10 released significant levels of plasma bovine prolactin
from at least as early as day 4 and continued to
release for at least 85 days, while formulation (b)
released significant levels from at least as early as
day 8 for at least 85 days.

What we claim is:-

1.   A pharmaceutical composition comprising a
polylactide, as hereinbefore defined, and an acid-stable
polypeptide, which, when placed in an aqueous
5   physiological-type environment, releases polypeptide
into said aqueous physiological-type environment in
a continuous manner, as hereinbefore defined, until
essentially all of the polypeptide has been released.

2.   A pharmaceutical composition comprising a
10   polylactide, as hereinbefore defined, and an acid-stable
polypeptide, and exhibiting two successive phases of
release of polypeptide when placed in an aqueous
physiological-type environment, the first phase being
released by matrix diffusion and the second phase being
15   released consequent upon degradation of the polylactide ,
characterised in that the diffusion phase and the
degradation-induced phase overlap in time.

3.   A pharmaceutical composition comprising a
polylactide as hereinbefore defined and an acid-stable
20   polypeptide, which, when placed in an aqueous physio-
logical-type environment absorbs water in a continuous
manner, as hereinbefore defined, until the polylactide
has been degraded and essentially all of the polypeptide
has been released into said aqueous physiological-
25   type environment.

4.   A pharmaceutical composition as claimed in
claim 1, 2 or 3 wherein the acid-stable polypeptide is
oxytocin, vasopressin, adrenocorticotrophic hormone
(ACTH), epidermal growth factor (EGF),
30   prolactin, luliberin or luteinizing hormone releasing

- 45 -

hormone (LH-RH), insulin, somatostatin, glucagon, interferon, gastrin, tetragastrin, pentagastrin, urogastrone, secretin, calcitonin, enkephalins, endorphins, angiotensins, renin, bradykinin, bacitracins, polymyxins, colistins, tyrocidin, gramicidines, and synthetic analgues and modifications and pharmacologically-active fragments thereof.

5. A pharmaceutical composition as claimed in claim 1, 2 or 3 wherein the polylactide has a high degree of heterogeneity, in respect of glycolide-rich and lactide-rich molecules, or of high polydispersity.

6. A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 5 to 50% by weight of a polypeptide of molecular weight less than 2000 and from 50 to 95% by weight of a polylactide wherein the ratio of glycolide to lactide units is from 0.5 to 3, and which has an inherent viscosity of greater than 0.5.

7. A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 5 to 50% by weight of a polypeptide of molecular weight less than 2000, and from 50 to 95% by weight of a polylactide wherein the ratio of glycolide to lactide units is from 0.2 to 3, and which has an inherent viscosity of 0.2 to 0.5.

8. A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 0.1 to 50% by weight of a polypeptide of molecular weight less than 2000 and from 50 to 99.9% by weight of a polylactide wherein the ratio of glycolide to lactide units is 0 to 3, and which has an inherent viscosity of less than 0.2.

9.   A pharmaceutical composition as claimed in
claim 1, 2 or 3 comprising from 10 to 50% by weight
of a polypeptide of molecular weight 1,500 to 10,000
and from 50 to 90% by weight of a polylactide wherein
the ratio of glycolide to lactide units is 0.5 to 3,
and which has an inherent viscosity of 0.4 to 0.8.

10.   A pharmaceutical composition as claimed in
claim 1, 2 or 3 comprising from 5 to 30% by weight
of a polypeptide of molecular weight 1,500 to 10,000
and from 70 to 95% by weight of a polylactide wherein
the ratio of glycolide to lactide units is 0.2 to 3,
and which has an inherent viscosity of 0.15 to 0.4.

11.   A pharmaceutical composition as claimed in
claim 1, 2 or 3 comprising from 0.1 to 20% by weight
of a polypeptide of molecular weight 1,500 to 10,000
and from 80 to 99.9% by weight of a polylactide wherein
the ratio of glycolide to lactide units is 0 to 3, and
which has an inherent viscosity of less than 0.15.

12.   A pharmaceutical composition as claimed in
claim 1, 2 or 3 comprising from 0.1 to 50% by weight
of a polypeptide of molecular weight 8,000 to 30,000
and from 50 to 99.9% by weight of a polylactide
wherein the ratio of glycolide to lactide units is 0 to
3, and which has an inherent viscosity of 0.15 to 0.4.

13.   A pharmaceutical composition as claimed in
claim 1, 2 or 3 comprising from 10 to 50% by weight of
a polypeptide of molecular weight 8,000 to 30,000 and
from 50 to 90% by weight of a polylactide wherein the
ratio of glycolide to lactide units is 0.7 to 3, and
which has an inherent viscosity of 0.1 to 0.15.

14.　　A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 0.1 to 50% by weight of a polypeptide of molecular weight 8,000 to 30,000 and from 50 to 99.9% by weight of a polylactide wherein the ratio of glycolide to lactide units is 0 to 3, and which has an inherent viscosity of less than 0.1.

15.　　A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 5 to 50% by weight of ICI.118,630 and from 50 to 95% by weight of a polylactide wherein the ratio of glycolide to lactide units is 0.8 to 3, and which has an inherent viscosity of more than 0.5.

16.　　A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 5 to 50% by weight of ICI.118,630 and from 50 to 95% by weight of a polylactide wherein the ratio of glycolide to lactide units is 0.2 to 3, and which has an inherent viscosity of 0.2 to 0.5.

17.　　A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 0.1 to 50% by weight of ICI.118,630 and from 50 to 99.9% by weight of a polylactide wherein the ratio of glycolide to lactide units is 0 to 3, and which has an inherent viscosity of less than 0.2.

18.　　A pharmaceutical composition as claimed in claim 1, 2 or 3 comprising from 10 to 50% by weight of epidermal growth factor or urogastrone and from 50 to 90% by weight of a polylactide wherein the ratio of glycolide to lactide units is 0.5 to 3, and which has an inherent viscosity of 0.4 to 0.8.

19.   A pharmaceutical composition as claimed in
claim 1, 2 or 3 comprising from 0.1 to 50% by weight
of epidermal growth factor or urocastrone and from
50 to 99.9% by weight of a polylactide wherein the
5   ratio of glycolide to lactide units is 0.3, and which
has an inherent viscosity cf 0.15 to 0.4.

20.   A pharmaceutical composition in the form of
a suspension for in injection comprising from   1   to
50   % by weight of a solid formulation, which itself
10   comprises from 0.1 to 50% by weight of an acid-stable
polypeptide as herein defined and from 50 to 99.9%
by weight of a polylactide hwerein the ratio of
glycolide to lactide units is 0 to 3 and which is either
soluble in benzene and has an inherent viscosity (1g./
15   100ml. solution in benzene) of less than 0.5 or is
insoluble in benzene and has an inherent viscosity (1g./
100ml. solution in chloroform or dioxan) of less than 4,
which solid formulation has been reduced to fine particle
size, together with from   50   to   99   % by weight of
20   a liquid carrier suitable for injection into mammals.

21.   A heterogeneous copolymer of lactic acid and
glycolic acid units comprising from 25 to 100% molar of
lactic acid units and from 0 to 75% molar of glycolic
acid units, and which is either soluble in benzene and
25   has an inherent viscosity (1g./100ml. solution in benzene)
of less than 0.5, or is insoluble in benzene and has an
inherent viscosity (1g./100ml. solution in chloroform or
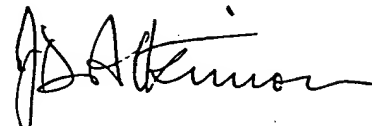dioxan) of less than 4.

22.   A process for the manufacture of a heterogeneous
30   copolymer as claimed in claim 21 which comprises the ring
opening copolymerisation of a mixture of the cyclic dimers
of lactic acid and glycolic acid.

- 49 -

23.   A heterogeneous copolymer as claimed in claim
21 whenever prepared by the process claimed in claim 22.

JOHN DAVID ATKINSON

AUTHORISED REPRESENTATIVE
General Authorisation No. 97

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl. 3)
D X Y	US - A - 3 773 919 (BOSWELL et al.) * examples 3, 6; column 8, lines 6 to 12 * * abstract; column 2, line 60, column 8, line 6 to column 11, line 49 *	21-23 1-20	A 61 K 37/02 C 08 G 63/08 /C 07 C 103/52 A 61 K 31/765 A 61 K 9/22
Y	Chemical Abstracts vol. 88, no. 12, March 1978 Columbus, Ohio USA T.M.S. CHANG "Biodegradable semiper- meable microcapsules containing enzymes, hormones, vaccines, and other biologicals" page 79044, column 2, abstract no 79041n & J. Bioeng. vol. 1, no. 1, 1976, pages 25 to 32	1-20	
	US - A - 3 886 699 (YOLLES) * claim 1; column 5, line 4; column 6, line 15 *	1-23	TECHNICAL FIELDS SEARCHED (Int. Cl. 3) A 61 K 9/22 A 61 K 31/765 A 61 K 37/02 C 08 G 63/08
	- A - 4 (?) 312 (SAUTER et al.) xamples *	21-23	
	A2 - 0 (?) (ELI LILLY AND CO.) ...ples 1 to 7 *	21-23	

./..

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
	10-05-1982	BREW

.005848

European Patent Office

EUROPEAN SEARCH REPORT

Application number

EP 82 30 0416.3
- page 2 -

	DOCUMENTS CONSIDERED TO BE RELEVANT		CLASSIFICATION OF THE APPLICATION (Int. Cl.³)
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	
A	Chemical Abstracts vol 91, no. 2, 1979 Columbus, Ohio, USA D. WISE et al. "Results on biodegradable cylindrical subdermal implants for fertility control" page 275, column 2, abstract no. 9442g & Midl. Macromol. Monogr., vol. 5 (Polym. Delivery Syst.), 1978, pages 121 to 138 --	21-23	
A	GB - A - 2 008 135 (ETHICON, INC.) * complete document * ----	21-23	**TECHNICAL FIELDS SEARCHED (Int. Cl.³)**